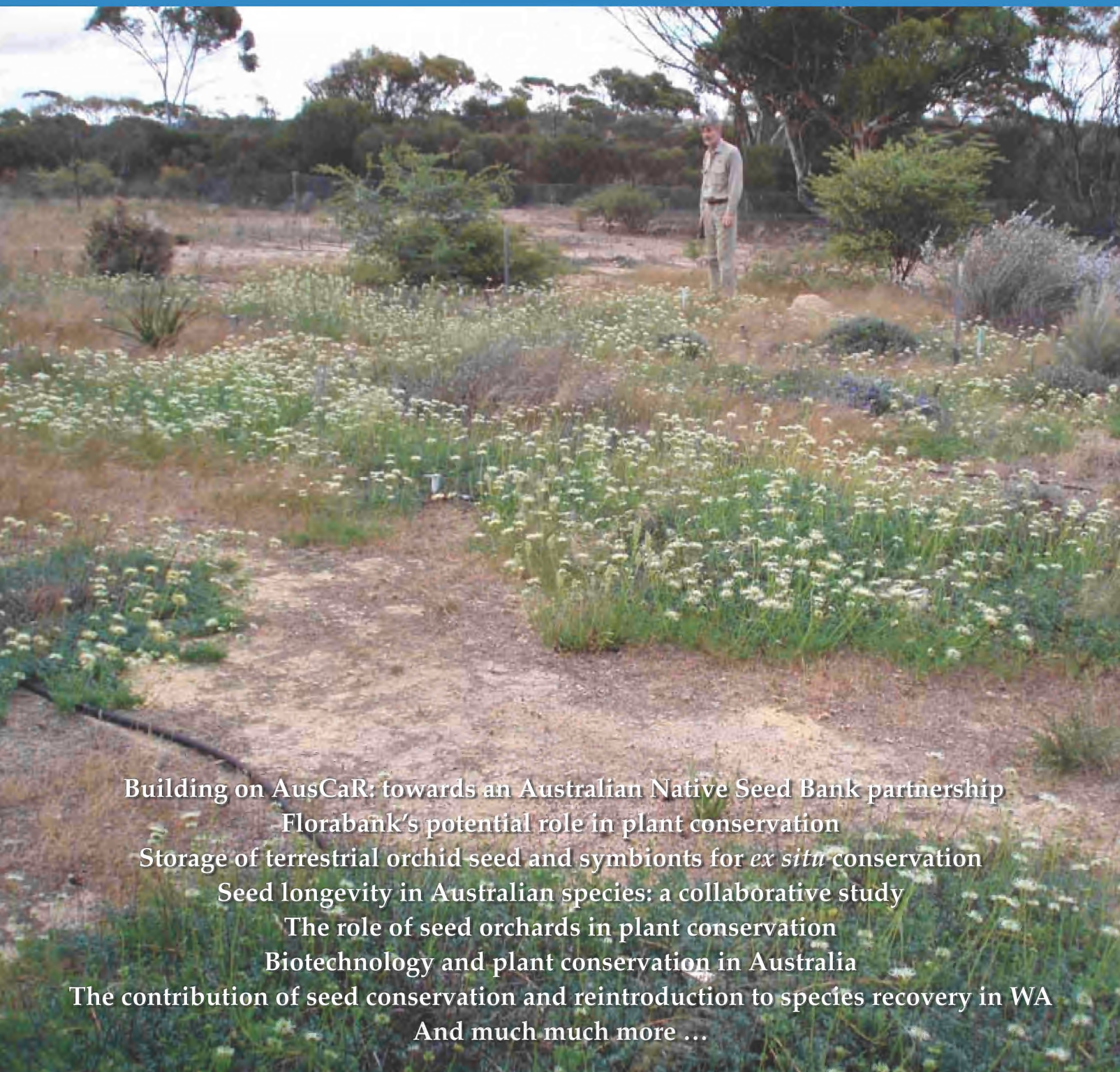


Australasian Plant Conservation

BULLETIN OF THE AUSTRALIAN NETWORK FOR PLANT CONSERVATION INC

VOLUME 17 NUMBER 4 • MARCH - MAY 2009



Building on AusCaR: towards an Australian Native Seed Bank partnership

Florabank's potential role in plant conservation

Storage of terrestrial orchid seed and symbionts for *ex situ* conservation

Seed longevity in Australian species: a collaborative study

The role of seed orchards in plant conservation

Biotechnology and plant conservation in Australia

The contribution of seed conservation and reintroduction to species recovery in WA

And much much more ...

SPECIAL THEME: GERMPLASM CONSERVATION—SAVING PLANTS FOR THE FUTURE

This issue

| | |
|---|----|
| Guest Editorial | 1 |
| A note from the President | 1 |
| Building on AusCaR: towards an Australian Native Seed Bank partnership | 2 |
| Florabank's potential role in plant conservation | 5 |
| Storage of terrestrial orchid seed and symbionts for <i>ex situ</i> conservation | 7 |
| Seed longevity in Australian species: a collaborative study through the AuSCaR (Australian Seed Conservation and Research) network | 9 |
| The role of seed orchards in plant conservation | 10 |
| Biotechnology and plant conservation in Australia: tissue culture and cryogenic research for <i>ex situ</i> conservation and restoration of endangered plants | 12 |
| The road to recovery: the contribution of seed conservation and reintroduction to species recovery in Western Australia | 15 |
| Black Gum, a nationally threatened tree of upland New South Wales and Victoria | 18 |
| A new population of Minyon Quandong (<i>Elaeocarpus sedentarius</i>) from northern New South Wales | 20 |
| Is fencing enough to conserve Western Australia's wheatbelt woodlands? | 22 |
| Endangered plant discovered by seven year old boy | 23 |

Regular Features

| | |
|---|----|
| Research Roundup | 24 |
| Information Resources and Useful Websites | 24 |
| ANPC Forum | 26 |
| Conferences and Workshops | 26 |

Australasian Plant Conservation

Editors

Phillip Ainsley and Rosemary Purdie

Editorial Team

Paul Adam, Phil Ainsley, Jim Crennan,
Thomas North and Zoe Smith

Layout & Graphic Design

Siobhan Duffy

Australasian Plant Conservation is produced by the ANPC Inc. with assistance from the Australian National Botanic Gardens.

Australasian Plant Conservation is printed on recycled paper.
ISSN 1039-6500

ANPC National Office

GPO Box 1777, Canberra, ACT 2601, Australia

Ph: (02) 6250 9509

Fax: (02) 6250 9528

Email: anpc@anpc.asn.au

Web: <http://www.anpc.asn.au>

ANPC Plant Conservation Email List

To subscribe, unsubscribe or post a message
send a request to anpc@anpc.asn.au

National Office Staff

Bruce Boyes, Sally Stephens
and Pamela Strickland

Volunteers

Merryl Bradley, Odette Mayne
and Richard Bomford

ANPC Committee

President

Bob Makinson

Vice President

David Coates

Treasurer

Vacant

Secretary

Helena Mills

Committee Members

Paul Adam, Phil Ainsley, Tom Celebrezze,
Paul Gibson Roy, Roger Good,
Deanna Marshall, Leonie Monks, Zoe Smith,
David Taylor and Judy West

New Zealand Plant Conservation Network

President Ian Spellerberg

Secretary John Sawyer

PO Box 5086 Wellington, New Zealand.

Email: info@nzpcn.org.nz



ANPC Inc.

Mission Statement

*"To promote and improve
plant conservation"*

Contributing to Australasian Plant Conservation

Australasian Plant Conservation is a forum for information exchange for all those involved in plant conservation: please use it to share your work with others. Articles, information snippets, details of new publications or research, and diary dates are welcome. **The deadline for the June–August 2009 issue is Friday 29 May 2009.** The theme of that issue will be 'Conservation on private land'. General articles are also very welcome. Please contact Rosemary Purdie if you are intending to submit an article: Rosemary.Purdie@environment.gov.au.

Authors are encouraged to submit images with articles or information. Please submit images as clear prints, slides, drawings, or in electronic format. Electronic images need to be at least 300 dpi resolution, submitted in at least the size that they are to be published, in tif, jpg or gif format. Guidelines for authors are at: <http://www.anpc.asn.au/anpc/pdf/APCGuideContrib.pdf>.

Please send articles, no more than 1100 words, as a MS Word (2000 compatible) or rich text format file, on disk or by email to: Rosemary.Purdie@environment.gov.au.

Opinions expressed in this publication are those of the authors and are not necessarily those of the ANPC or its sponsors. Material presented in *Australasian Plant Conservation* may be copied for personal use or published for educational purposes, provided that any extracts are fully acknowledged. Where any material is credited to and/or copyright to another source, please contact the original source for permission to reprint.

Front cover: Plants of the critically endangered Corrigin Grevillea (*Grevillea scapigera*) thriving in a re-introduced population. Photo: I.R. Dixon
Cover design: Siobhan Duffy.
Printed by: Goanna Print, Canberra.

Guest Editorial

Germplasm conservation – saving plants for the future

Phillip Ainsley

Botanic Gardens of Adelaide, SA. Email: ainsley.phillip@saugov.sa.gov.au

Over the last decade the global crisis associated with species extinctions has become one of the key priorities for conservation programs around the world. Paralleling this has been the rapid uptake and increased adoption of *ex situ* germplasm conservation strategies to complement on ground *in situ* activities in the battle to stem the loss of plant species and their genetic diversity.

Almost 12 years ago the Australian Network for Plant Conservation (ANPC) produced the first version of the publication *Germplasm Conservation Guidelines for Australia*. The thinking behind this document was to provide an introduction to the principles and practices for seed and germplasm banking that researchers and practitioners could apply to Australian native plant species.

Whilst the publication has served us well, the technologies available and knowledge relating to germplasm conservation have greatly increased and improved. It therefore seems an appropriate time to revise this publication and ensure that information about the most current thinking and relevant methodologies are available to all involved in germplasm conservation. Over the last 12 months researchers and practitioners around Australia have been working to revise the ageing document, and it is anticipated that a revised edition will be launched later this year.

As a glimpse of what to expect, we have dedicated this edition of *Australasian Plant Conservation* to 'Germplasm Conservation' and the role it is playing in saving plants for the future in Australia. Contributions have been made from around the country.

The articles start with an overview about the current seed bank conservation activities in Australia and an update about the achievements of partners affiliated with the AuSCaR (Australian Seed Conservation and Research) network (Tom North). This is followed by an update about the FloraBank Project (Penny Atkinson). These two articles highlight the commitment and extent of germplasm conservation activities occurring around Australia, and the contribution that we are making to the global effort to combat species loss.

Articles then focus on methods and emerging techniques associated with germplasm conservation. They include an overview of orchid *ex situ* conservation (John Siemon, Cathy Offord and Karen Sommerville), research that is underway to understand the longevity of seeds from Australian native plants (Amelia Martyn), the role of seed orchards (Anne Cochrane and Sarah Barrett) and using biotechnology techniques such as plant tissue culture and cryopreservation (Eric Bunn and Shane Turner) in plant conservation. The final germplasm conservation article is a case study about the contribution of seed conservation and reintroduction to species recovery in Western Australia (Andrew Crawford and Leonie Monks).

In the rest of the issue you can read about the Black Gum, a nationally threatened tree from upland New South Wales and Victoria (Steve Douglas), the discovery of a new population of Minyon Quandong in northern New South Wales (Lui Weber) and learn about a study to determine how successful fencing has been for conserving Western Australia's wheatbelt woodlands (Rachel Standish, Suzanne Prober, Chris Curnow and Jeff Richardson).

Happy reading!

ANPC OFFICE: THANK YOU SALLY, WELCOME BRUCE

Sally Stephens finished work as ANPC's Project Manager in early March after five years in the job. Over that time she has been responsible for organising a wide range of highly successful ANPC workshops, greatly extending our network, building our partnerships and helping make sure we meet our key strategic objective of facilitating the exchange of high quality plant conservation information between scientists and on-ground practitioners. A big thank you to Sally for all the energy, creativity, patience and good humour she has brought to the position.

With Sally's departure we welcome our new Project Manager, Bruce Boyes, who joins us from Queensland via New South Wales and the ACT. Bruce has a strong background and an extensive network in natural resource management across Australia, lots of experience in making grant applications and coordinating and organising conferences and workshops, and has also been active in a range of knowledge transfer and management programs. We warmly welcome Bruce to our ANPC office in Canberra, and look forward to working with him.

Bob Makinson, ANPC President

Building on AuSCaR: towards an Australian Native Seed Bank partnership

Tom North

*Australian Seed Conservation and Research (Millennium Seed Bank–Australian Partners),
c/- Kings Park and Botanic Garden, West Perth, WA. Email: thomas.north@bgpa.wa.gov.au*

Introduction

The Millennium Seed Bank (MSB) – Australian partners, also known as Australian Seed Conservation and Research (AuSCaR), have been working together since 2005 to help conserve Australia's plant life. The network has largely been facilitated and funded as part of Royal Botanic Gardens, Kew's Millennium Seed Bank Project (MSBP).

In developing and maintaining conservation seed banks, AuSCaR has been dealing in low volumes of native plant seed for long-term storage of high value endemic, rare, threatened or significant species. Stocks have been utilised for research to resolve problems relating to storage, germination and delivery of seed; these may be species-specific or generic across families. Results are and have been applied to improve the efficiency of, and the suite of species included in, revegetation / restoration projects. The seed banks are all involved in aiding translocations and acting as a supplementary tool for recovery plans of threatened species.

At a national level, AuSCaR has provided a network of capacity building, training and collaborative scientific research opportunities that its members can tap into. At an international level, AuSCaR partners can share knowledge and expertise with a global network of up to 123 organisations from the 54 countries that make up Kew's MSBP.

The network has been working towards the achievement of Target 8 of the *Global Strategy for Plant Conservation* by 2010: '60 per cent of threatened plant species in accessible *ex situ* collections, preferably in the country of origin, and 10 per cent of them included in recovery and restoration programs'.

Persistent drought and the inherent nature of collecting from rare or threatened species have impacted on collection rates. Drought has restricted collections from target species throughout the program, with most partners attempting collections under the most severe conditions on record.

Review of Progress

At present seed from 21% of Australia's threatened plant species is in storage, of which at least 10% have been used for recovery and restoration. By the end of 2009, AuSCaR partners aim to have collected and stored seed from around a third of Australia's flora (about 8,000 species).



Representatives of the AuSCaR network at a meeting held in Canberra in 2008. Photo: James Wood

The AuSCaR partners provide seeds for duplicate storage at Kew's Millennium Seed Bank in the UK. AuSCaR is making a significant contribution towards Kew's MSBP target of collecting and conserving 10% of the world's flora by 2010.

The next phase of Kew's MSBP, from 2010 to 2020, will see 25% of the world's flora secured in long-term storage. Kew's MSBP also hopes to embark on an ambitious program of species reintroduction and restoring damaged habitats. This is an area in which AuSCaR's partners—particularly in New South Wales, South Australia and Western Australia—are leading the world, and have valuable experience to share both within Australia and with the rest of the MSBP's member countries.

Kew's Millennium Seed Bank Partnership is at a critical stage in its life, with further funding (Aus\$230 million) needed in order to continue developing its vital work and achieving its 2020 aims. AuSCaR itself has set a goal of raising \$29 million to help implement the new national partnership.

AuSCaR: Member Activities (states in alphabetical order)

New South Wales

Partner: NSW Seedbank, Royal Botanic Gardens Trust, Sydney.

'SeedQuest NSW' was launched in 2003. Five years on, seed from a third of the state's flora is being kept safe from

threats such as climate change in the NSW Seedbank and Kew's Millennium Seed Bank in the UK.

One third of New South Wales' threatened plant species are now held, and in 2008 the 1000th SeedQuest seed collection (of the Downy wattle (*Acacia pubescens*), a listed Vulnerable species) was achieved. Seed research has increased during the project and includes seed dormancy in groups such as Rutaceae, and accelerated seed ageing. Seed batches were recently sent on a NASA space shuttle flight for short-term storage on the International Space Station. They included Golden Wattle (*Acacia pycnantha*), Waratah (*Telopea speciosissima*), Flannel Flower (*Actinotis helianthi*) and Wollemi Pine (*Wollemia nobilis*).

The NSW Seedbank last year launched the 'Australian Rainforest Seed Project' which is investigating the biology and conservation of rainforest species, whose seeds are sensitive to drying out and not usually suitable for seed banking.

More information:

<http://www.rbgsyd.nsw.gov.au/welcome_to_bgt/feature_stories/nsw_seedbank2/seedquest_nsw>

Northern Territory

Partner: The Department of Natural Resources, Environment, The Arts and Sport (NRETAS) of the Northern Territory Government.

The Kew partnership with the NT seed project began at the end of 2004, with a total collection target of 550 new species to Kew by the end of 2010. At the end of the fourth year, seed from approximately 370 new species had been collected for Kew. An additional 200 duplicate species have also been conserved in the NT Seed Bank.

Approximately 70 of the existing collections are considered priority collections due to the species being endemic, rare or threatened at the national level (e.g. *Ipomoea polpha* subsp. *latzii*, *Eremophila* sp. Rainbow Valley) and state and/or regional levels. Other species have been collected due to their indigenous or biological importance (e.g. *Terminalia carpentariae* as a food source for the Vulnerable Arnhem Rock Rat (*Zyzomys maini*)).

Collecting efforts have also involved working with aboriginal communities, including rangers from the Docker River Community. In 2007, the Docker River rangers assisted in collecting approximately 25 species from their lands (Petermann Aboriginal Land Trust), including the Northern Territory endemic *Rulingia luteiflora*.

More information:

<<http://www.nt.gov.au/nreta/>>

Queensland

Partners: Brisbane Botanic Gardens; University of Queensland; Griffith University; Greening Australia, Queensland; Environmental Protection Agency; Australian Centre for Mining and Engineering Research.

Queensland has the second highest number of threatened species in Australia, and the 'Seeds for Life' project is collecting and protecting seeds from 1000 of Queensland's most vulnerable plant taxa. To date they have collected over 600 species new to the Millennium Seed Bank.

Griffith University is a collaborating partner with 'SeedQuest NSW' in investigating the issues surrounding the seed banking of rainforest species, under Royal Botanic Gardens Foundation funding. The University of Queensland has had joint funding with the Western Australian Botanic Gardens and Parks Authority (BGPA) to investigate issues surrounding the use of native seed in mine site restoration.

More information:

<<http://www.greeningaustralia.org.au>>

South Australia

Partner: Botanic Gardens of Adelaide, Department for Environment and Heritage.

The SACRED Seeds (South Australian Collection of Rare and Endangered Seeds) project is based at the Botanic Gardens of Adelaide Seed Conservation Centre. Since the project commenced in 2003, seeds have been collected from more than 1500 (42%) of the state's native plant species, including over 400 (50%) of its listed threatened taxa.

The Botanic Gardens of Adelaide was the first partner to the MSBP to meet its collection target, and in December 2008 the 1010th species considered new to the collections of the Millennium Seed Bank was handed over for duplicate storage in the UK. The project has provided an opportunity to rediscover species that were believed to be extinct at both state and regional levels (*Cullen microcephalum*, *Oreomyrrhis eriopoda*, *Viola betonicifolia* subsp. *betonicifolia*). It has also partnered with external stakeholders to provide seedling material from 32 species for inclusion in regional restoration and rehabilitation projects.

More information:

<http://www.environment.sa.gov.au/botanicgardens/seed_conservation.html>

Tasmania

Partners: Nature Conservation Branch, Department of Primary Industries and Water; Royal Tasmanian Botanic Gardens and Tasmanian Herbarium.

Tasmania became part of Kew's MSBP in 2004. Its partners aim to collect seed from 800 species, which is close to half the total Tasmanian flora. The Tasmanian team recently had their most successful field trip, collecting 82 species over 8 days on the Central Plateau.

The team is also having success with one of the state's rarest species, Davies' Waxflower (*Phebalium daviesii*), for which only 23 plants exist in the wild. Through a simple change to collection techniques, the Royal Tasmanian Botanic Gardens' seed collection of Davies'

Waxflower was boosted to 34,500 seeds in January 2009. A collection of this size will enable the Gardens to gain an understanding of the conditions and factors required for the reproduction of this difficult-to-germinate species. Once this is understood, the 34,500 seeds will have real value in the species' long-term conservation, both in controlled conditions at the Royal Tasmanian Botanic Gardens and hopefully in the wild.

More information:

<http://www.rtbg.tas.gov.au/tas_seed_conservation.html>

Victoria

Partners: The National Herbarium of Victoria; Department of Sustainability and Environment.

The Victorian Conservation Seedbank has a particular focus on some 450 of the state's endemic and most threatened flora. They have played a key role in the delivery of the Victorian Threatened Orchid Recovery Project, which covered 80 species, saving four Critically Endangered orchid species from extinction.

The Victorian Conservation Seed Bank has also played a major part in the recovery plan for the 'Presumed Extinct' Wimmera Rice-flower (*Pimelea spinescens* subsp. *pubiflora*).

More information:

<http://www.rbg.vic.gov.au/research_and_conservation/seedbank>

Western Australia

Partners: Department of Environment and Conservation (DEC); Botanic Gardens and Parks Authority (BGPA).

Western Australia has been working with Kew since 2001 and is the original Australian partner in Kew's MSBP. By 2010 the Western Australia partners will have conserved seed from 2,500 native species in the state. This total already includes 70% of the state's threatened flora and more than 10% will have been used in recovery programs.

Seedlings of the Critically Endangered Brown's Banksia (*Banksia brownii*) derived from a climate change seed conservation research project between DEC and Kew's MSBP scientists were planted out in the wild in 2008. Many of Australia's banksias are threatened in the wild due to the fungal dieback disease *Phytophthora cinnamomi*. The seedlings were found to be genetically distinct from the remaining wild populations of *B. brownii*, highlighting just how vital seed collection and conservation is when plant conservationists cannot adequately maintain genetic diversity of species in the wild in the long-term.

More information:

<<https://www.dec.wa.gov.au/science-and-research/plant-research/seed-banking-for-biodiversity-conservation/the-threatened-flora-seed-centre.html>>

<<http://www.bgpa.wa.gov.au/o/content/view/241>>

The Future: a New Partnership

The launch of the *National Strategy and Action Plan for the Role of Australia's Botanic Gardens in Adapting to Climate Change* (the National Strategy) (<<http://www.anbg.gov.au/anbg/botclimate/index.html>>) and its subsequent endorsement by the Natural Resource Management Ministerial Council has given new impetus to the work of Australia's conservation seed banks. In December 2008, the lead agencies and organisations involved in seed banking met in Canberra to develop a coordinated strategy aimed at delivering the first goal, 'A national safety net for Australia's plant species' over the next ten years (2010-2019).

Seventeen institutions and organisations are now in the process of initiating a new partnership, the 'Australian Seed Bank'. The organisations involved at this stage consist of AuSCaR members, the Australian National Botanic Gardens and Greening Australia's Florabank, although it is hoped that the partnership will grow. This new partnership is recognition of the need not only to develop a nationally coordinated approach to seed banking and research, but also to improve integration of conservation seed banking with on-ground landscape restoration activities and priorities.

In addition to the need to safeguard genetic material in conservation seed banks, future availability, supply and management of seed will also be significant challenges for biodiversity management at larger landscape scales. The partners between them have the skills and knowledge to help tackle these challenges. Through the broader perspective of the new partnership it is hoped that the important work of AuSCaR will form the building blocks for a new unified vision supporting plant conservation.

Florabank's potential role in plant conservation

Penny Atkinson

Florabank, Greening Australia, Canberra, ACT. Email: PAtkinson@greeningaustralia.org.au

Around Australia, hundreds of tonnes of native seed are traded for restoration and remediation projects in mining, natural resource management, and for use in the national, state and local government sectors. Florabank is about improving the results of these projects through improving the availability of appropriately sourced, good quality seed for large-scale restoration projects.

At the moment, the supply of native seed is not meeting demand in many places. This compromises restoration work either by reducing the extent of projects, or by limiting them to species where seed or tubestock is easy to obtain and thus compromising the biodiversity values of projects. Seed supplied is often of poor quality, which again lowers the success and biodiversity of restoration projects. There is a need to improve both the availability and quality of seed, and Florabank is trying to meet this need by working with the native seed sector and the research community. We work to improve information access, have developed training, and are working towards industry accreditation and certification for better restoration outcomes.

Florabank's focus has always been on using local seed banks to supply seed for successful restoration, in the quantities and taxa required for community, public and commercial restoration projects. In this way, it complements the work done by the Australian Seed Conservation and Research Network (AuSCaR), and by organisations such as the Australian National Botanic Gardens (ANBG) which have predominantly focused on threatened species and smaller scale projects. We would now like to work to

better incorporate the restoration of threatened species and threatened communities into conventional restoration projects through a new national partnership between AuSCaR, the Australian Network for Plant Conservation (ANPC), ANBG and Florabank.

About Florabank

Florabank formed in 1998 as a partnership between Greening Australia, ANBG and CSIRO's Australian Tree Seed Centre (then in the Division of Forestry and Forest Products, now in CSIRO Plant Industry). The initial project began with extensive consultation, visits to seed banks around Australia, and two national surveys of people involved in seed and restoration in the commercial, government and community sectors. Florabank continued from there to become a respected provider of information to the on-ground native seed sector. It was realised that much of the crucial knowledge about native seed is hidden in scientific papers and reports (which are not accessible to people working on the ground), or else, hidden in people's heads because they have been working with seed for decades but do not have the means to widely share that knowledge.

Our Website

The Florabank website (<http://www.florabank.org.au/>) is a popular and well-used site, and an excellent means of communicating with the native seed sector. It includes a Registered Users facility, which currently has over 650 registered users, and information forums where practitioners ask and answer questions about native seed (for example, how to germinate *Macrozamia* or *Leucopogon* seed). We hope to use 'citizen science' to enable better transfer of knowledge between researchers and the people who work with native seed across Australia. The website also contains the following tools.

- The **Species Navigator** assists people to identify the right species for their restoration project, and provides them with information about how to collect, plant and use these species. Landholders and project managers can use this tool to select species suitable for different planting purposes (e.g. saline sites, wood products, biodiversity, etc) so that appropriate species that will grow in specific site conditions can be selected and used. The **Site Description Tool** works with the Species Navigator and assists people to find out and record the right information about their site. This will enable them to fully use the Species Navigator, or to work with their local seed supplier or nursery to select the most appropriate species for their project.



Florabank training participants visit a seed production area, SA. Photo: Paul Macdonell

- The **Seed Collection Advisor** assists people to make the right decisions in order to collect seed with a good genetic base. It is a decision support tool which steps users through considering their local landscape, the biology of the species, and the population distribution, and then ends with recommendations to improve the genetic diversity of collected seed.
- The **Vegetation Management Tool** assists people to plan, implement, maintain and monitor their restoration sites. This is set out like an online book, but with embedded links to further information, references and other resources. This tool can be updated with new information, examples and references.

These tools were co-developed with CSIRO. Florabank plans to build on them over the next five years so they cover more species and include new research. Our aim is to include 600 taxa in Species Navigator, which would then provide information on over 80% of commonly used restoration species.

Our Publications

Florabank has published a range of documents for the native seed sector in Australia, including the *Model Code of Practice for Community Based Collectors and Suppliers of Native Seed* and ten *Florabank Guidelines* (1999-2000). The guidelines have been the main reference for people using native seed in the commercial, government and community sectors and have resulted in the development and success of hundreds of local community seed banks and nursery projects.

Florabank has also produced technical articles, and initiated events drawing on native seed science and technical expert sources. This work is continuing: currently we are working with a scientific panel led by CSIRO on a revision of the *Florabank Guidelines* on Provenance, and also working towards revision of the Seed Production Areas guideline. The results of these reviews will be incorporated into the *Florabank Guidelines* and the Seed Collection Advisor, industry training, and industry codes of practice.

Supporting Professional Development

In 2007-08 Florabank worked with CSIRO to develop and deliver Florabank Professional Development Training. This is accredited at Certificate III level, and was designed for commercial and agency seed operators who collect and supply seed for restoration projects. Many people working in the seed sector have no formal training but have learnt 'on the job'. This course aims to provide them with an understanding of recent developments in seed science, including provenance, seed production areas and seed supply planning.

This practically-focused course was successfully delivered in six locations around Australia, with feedback showing



Native seed for restoration in storage, ACT.
Photo: Kimberlie Rawlings

87% of respondents used the information to change their workplace practices. Even experienced and respected operators who had been working in the sector for 10-15 years found the course hugely beneficial. Florabank is continuing the delivery of this course, and is now working towards the establishment of a new Certificate I native seed unit with Agrifood Skills Australia.

Conclusion

By working with native seed researchers, and making information available about better practices in native seed collection, handling and establishment, we hope that more species will be used in restoration projects, and that future projects will better represent the local plant communities and incorporate threatened species and communities. Communicating the results of native seed and restoration research directly to the native plant practitioner community through Florabank will have immense benefits for restoration success.

Storage of terrestrial orchid seed and symbionts for *ex situ* conservation

John Siemon, Cathy Offord and Karen Sommerville

Botanic Gardens Trust, Mount Annan Botanic Garden, NSW. Email: John.Siemon@rbgsyd.nsw.gov.au

Introduction

For many plant species, successful *ex situ* conservation is achieved through the long-term storage of their seeds. The storage of terrestrial orchid seeds, however, is complicated by their subsequent germination requirements. In natural environments, terrestrial orchid seeds require a mycorrhizal association (with symbiotic fungi) to supply the nutrients necessary for germination; in some cases, this association is highly specific. Arguably, then, the storage of these seeds for conservation purposes is not very useful without also isolating and storing the appropriate fungal symbiont/s.

Various tissue culture mediums are now available that allow the germination of orchid seeds without the presence of a fungal symbiont, as each medium is formulated with all the nutrients necessary to initiate germination and promote growth. However, if such seedlings were to be used for translocation purposes, the resulting populations would not be self-sustaining unless the specific mycorrhizal symbiont required for germination was present at the translocation site. This paper addresses the methods currently in use for storing both the seeds and the fungal symbionts of terrestrial orchids.

Seed Production and Collection

Orchid seed suitable for storage can be derived from open- or hand-pollinated plants, located either *in situ* or in *ex situ* collections. Seed pods can take from a few weeks to several months after pollination to mature, although in hot, dry periods, fully developed pods can mature in a matter of days, and the majority of dust-like seed may be dispersed within minutes.

One of the advantages of *ex situ* derived seed is that pod maturation may be monitored more closely, minimising the chance of seeds dispersing prior to pod collection. In the field, such losses may be minimised by covering maturing pods with fine mesh bags (empty tea bags work well) when frequent visits to the site are not feasible. Alternatively, flower stems may be cut and kept in a vase of water until the pod matures (indicated by a change in colour from green to yellow or brown).

If the collected seed is to be introduced immediately to tissue culture, then the surface of fully mature pods may be sterilised and the pod opened under aseptic conditions to remove the seed. If the seed is to be kept for later use, the pods may be placed in a paper bag or envelope and stored in a cool, dry environment until the pods dehisce and the

seed is released naturally. Although other containers may be used, the electrostatic forces present in some plastic, glass or metal containers can make it difficult to remove the seed from the container. This is a particular problem when the species is threatened and seed numbers are limited.

Seed Drying and Storage

It has been common practice to dry and store orchid seed under ambient conditions. However, as viability may decline rapidly under these conditions, the method is useful only for short-term storage. Long-term storage is achieved most effectively under cool, dry conditions. Thus, orchid seed to be stored long-term should first be dried to approximately 5-6% moisture content. At Mount Annan Botanic Garden this is accomplished by placing the seed in a paper bag and holding it in a drying room with low temperatures (16-18°C) and low humidity (~16% relative humidity) for several weeks. For those not lucky enough to have a drying room, the seed can be dried in an airtight jar



Maturing pod of *Diuris arenaria*. Photo: John Siemon



Germinating *Pterostylis saxicola* seed in the laboratory. The seed was encapsulated in an alginate bead with a fungal symbiont. Photo: John Siemon

with a layer of silica gel placed at its bottom. Once dried, the seed should be sealed into an airtight packet (to prevent rehydration) and stored at low temperatures (see also next article by Amelia Martyn).

Many orchid growers store dried seeds in the crisper of a refrigerator at 4°C. Whilst refrigeration extends viability in comparison to storage at room temperature, it is still only a short-term storage solution. Long-term storage of dried orchid seeds is likely to be best achieved at sub-zero temperatures. For most growers, the only practical way to achieve this would be to store orchid seed in an ordinary freezer at -18°C, although even at this temperature seed may be relatively short-lived. If facilities are available, and the seed is dried properly, storage at -180°C and -196°C (in liquid nitrogen) has the potential to give greater longevity. Immersion of most seeds and fungal material in liquid nitrogen appears to have no deleterious effects. In some instances immersion of dried seed in liquid nitrogen has increased subsequent germination percentages (Batty *et al.* 2001).

An essential component of long-term seed storage is the periodic testing of seed viability. This may be accomplished through germination in the presence of the correct fungal symbiont, or on specially formulated asymbiotic media. Theoretically, orchid seed viability may also be detected using biological stains such as fluorescein diacetate (FDA), tetrazolium (TTZ) or Evan's blue. In some instances, however, the use of stains has been found to be a poor predictor of viability for terrestrial orchids (Batty *et al.* 2001).

Alternative Storage Methods

An alternative technique for conserving orchid seed is to simultaneously store both seed and the mycorrhizal symbiont/s in an alginate bead. In this technique, developed by Wood *et al.* (2000), the seed and fungus are mixed together in a solution of sodium alginate, then pipetted drop-by-drop into a calcium chloride solution to form individual beads. The beads are then soaked in a sucrose solution prior to dehydration and storage.

At Mount Annan Botanic Garden this technique has been successfully tested on two threatened orchids from NSW—*Diuris arenaria* and *Pterostylis saxicola* (Sommerville *et al.* 2008)—and is now being tested on three additional species. To date, it appears that the encapsulation-dehydration protocol is very effective and enables simultaneous storage of seed and fungal symbionts without loss of viability in either. This has the potential to greatly improve seed banking procedures for terrestrial orchids, reducing the need for maintaining dual collections and reducing the need for regular subculturing of the fungal symbionts. The use of this technique for translocation of threatened species is currently being investigated.

Conclusion

The germinability and storability of many Australian native plant species has now been documented but there is still a great deal we need to understand about our terrestrial orchids. Information on orchid seed storage contributes to both recovery planning and long-term storage protocols for native orchid species.

References

- Batty, A.L., Dixon, K.W., Brundrett, M. and Sivasithamparam, K. (2001). Long-term storage of mycorrhizal fungi and seed as a tool for the conservation of endangered Western Australian terrestrial orchids. *Australian Journal of Botany* 49: 619–28.
- Sommerville, K.D., Siemon, J.P., Wood, C.B. and Offord, C.A. (2008). Simultaneous encapsulation of seed and mycorrhizal fungi for long-term storage and propagation of terrestrial orchids. *Australian Journal of Botany* 56: 609–15.
- Wood, C.B., Pritchard, H.W. and Miller, A.P. (2000). Simultaneous preservation of orchid seed and its fungal symbiont using encapsulation-dehydration is dependent on moisture content and storage temperature. *Cryo Letters* 21: 125–36.

Seed longevity in Australian species: a collaborative study through the AuSCaR (Australian Seed Conservation and Research) network

Amelia Martyn

NSW Seedbank, Mount Annan Botanic Garden, NSW. Email: Amelia.Martyn@rbgsyd.nsw.gov.au

How Old?

Seeds of many Australian plants are expected to be long-lived in storage, with groups such as acacias, eucalypts and casuarinas topping the list. The long lifespan expected for seed of many Australian species is illustrated in New South Wales by a small batch of Golden Wattle (*Acacia pycnantha*) seed originating from Australia's first Arbor Day in 1890, where 80% of the seed germinated in 1990 after being stored for 100 years in a sock drawer!

However, the longevity of many species is unknown, so the Australian Seed Conservation and Research (AuSCaR) network (see www.auscar.org.au), the Australian partners of the Millennium Seed Bank, are working to rank a wide range of species from shortest to longest lived. This will determine which species are likely to survive for long periods in storage and which will need to be replenished regularly with fresh seed. The ranking is also great for prioritising which species must be cleaned and stored first—a key task at the end of a busy collection season.

Project Snapshot

Using a standardised rapid ageing protocol (Figure 1), scientists at Mount Annan Botanic Garden in New South Wales, Kings Park and Botanic Garden in Western Australia, the Botanic Gardens of Adelaide in South Australia, Griffith University in Queensland and the Millennium Seed Bank in the UK are working together to produce a 'snapshot' of Australian seed lifespan in storage. The rapid ageing technique for studying seed longevity is outlined at http://www.kew.org/msbp/scitech/publications/comparative_longevity.pdf.

Seed longevity is primarily influenced by moisture and temperature, with cool dry storage reducing seed ageing for most orthodox species (i.e. species that can withstand drying, termed desiccation tolerant species). Drying can be achieved with purpose-built low humidity drying rooms (Figure 2), cabinet dehumidifiers, sealed containers with seeds dried over silica gel, air conditioned rooms, sheds with drying racks, polythene igloos or ambient conditions in sun or shade. Cold storage includes household refrigerators, commercial deep freezers, purpose-built cold rooms, or storage at very low temperature (-196°C) in liquid nitrogen (cryostorage).

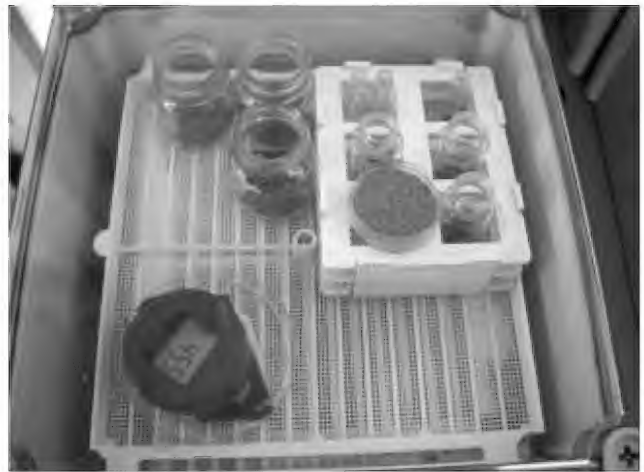


Figure 1. Rapid ageing experiment to determine seed longevity. Seeds are kept in a sealed box at 45°C and 60% relative humidity, with jars of seed removed at regular intervals and the seed germinated to monitor seed ageing over time. Photo: A. Martyn



Figure 2. Drying room of NSW Seedbank, where seeds are dried to 15% relative humidity at 15°C. Cool, dry storage is essential for increasing the longevity of seeds of orthodox species. Photo: Botanic Gardens Trust

Conservation Seedbanks

Conservation seedbanks, maintained by the AuSCaR partners, dry seeds at about 15% relative humidity and 15°C to reduce seed moisture content to 3-7%. Seeds are sealed into packets or airtight jars and stored at -18°C. These conditions are suitable for many Australian species (Offord *et al.* 2004; Crawford *et al.* 2007). Other cool, dry conditions may be more practical for restoration or nursery seed banks, depending on how quickly seed will be utilised.

References and Further Reading

Crawford, A.D., Steadman, K.J., Plummer, J.A. Cochrane, A. and Probert, R.J. (2007). Analysis of seed-bank data confirms suitability of international seed-storage standards for the Australian flora. *Australian Journal of Botany* 55:18–29.

Millenium Seed Bank Project (undated) *How long can seeds live?* <<http://www.kew.org/msbp/faq/longdoseedslive.htm>>.

Offord, C.A., Mckensy, M.L. and Cuneo, P.V. (2004). Critical review of threatened species collections in the NSW Seedbank: implications for *ex situ* conservation of biodiversity. *Pacific Conservation Biology* 10(4): 221–36.

Pain, S. (23 September 2006). Revival of the fittest. *New Scientist* 191(2750): 52–3.

The role of seed orchards in plant conservation

Anne Cochrane and Sarah Barrett

Department of Environment and Conservation, Albany, WA. Email: anne.cochrane@dec.wa.gov.au

Introduction

Seed orchards are a common method of mass-multiplication of plants for transferring genetically improved material from a breeding population to a plantation. They are often referred to as ‘multiplication’ populations. A seed orchard is often composed of selected genotypes derived from vegetative propagation, but seedling seed orchards also occur mainly to combine an orchard with progeny testing. Seed orchards are a strong link between breeding programs and plantation establishment. They are designed and managed to produce large and sustainable crops of seeds of superior genetic quality compared to those obtained from unmanaged wild stands. Seed orchards or seed production areas have been a traditional practice of forestry, where selection of material for propagation is made on desired traits or characteristics.

Seed Orchards for Conservation

More recently seed orchards have been established to assist in the conservation of critically threatened plant species. Plants in the orchard are more closely related to the parent population, and these plantings are aimed at producing material for reintroduction, reinforcement, habitat restoration and management. Genotypes for seed orchard planting are not selected for particular traits but are derived from as many wild plants as possible to retain the genetic diversity of the sourced population. In reality, these plantings should be termed ‘conservation seed orchards’ or ‘field genebanks’ in order to separate them from the traditional ‘seed orchard’ with their connection to genotype selection.

Threatened species that do not readily produce seeds or are highly threatened in the wild can be conserved in conservation seed orchards to maintain secure *ex situ* field populations and to facilitate the utilisation of this material in conservation programs. This occurs in particular when an *in situ* population is degraded or facing decline and eventual extinction, and where management intervention is largely unsuccessful. These conservation plantings are subject to intensive management which may include fertilisation, watering and herbivore protection, in addition to demographic monitoring. First generation propagated plants can be returned to the original *in situ* site to augment the wild population.

Advantages and Disadvantages

Conservation seed orchards are gaining acceptance as an appropriate strategy for the *ex situ* conservation of threatened species. They support the survival of wild populations in the same way that *ex situ* banking of seeds supports *in situ* conservation through provision of material for reintroduction and restoration and as a safeguard against extinction in the wild. A conservation seed orchard should be sufficiently large to maintain the genetic diversity of a population or species, and produce sufficient seeds for the establishment of self-sustaining populations in the wild. Use of seeds from a conservation seed orchard takes pressure off wild populations. These orchards are generally established for the production of woody perennials, however, with better understanding and cultural practices, herbaceous and grass species could be planted.

Conservation seed orchards have some disadvantages. These include the potential for hybridisation from closely-related taxa growing nearby and the possibility

that natural pollinators may be absent or in low number, thereby reducing the potential for seed set in species requiring cross fertilisation (outcrossing species). In the former case, plantings should be planned to avoid the risk of hybridisation occurring. In the latter case, appropriate artificial pollination strategies may need to be developed, requiring an understanding of the reproductive biology of the species. Conservation seed orchards can therefore provide interesting sites for research projects that aim to gain greater understanding of the targeted species. These findings can help support the conservation and management of wild populations.

The following example from the Department of Environment and Conservation (DEC) in Western Australia demonstrates the use of a seed orchard that supports the conservation of a highly threatened plant species in that state which faces an uncertain future.

Conservation Seed Orchard Case Study

Banksia montana (Figure 1) is one of a number of highly threatened narrow range endemics that inhabit the floristically diverse mountain peaks of the Stirling Range National Park in southern Western Australia. This species has faced reductions in population size and health in recent years due to a combination of frequent fire, grazing and dieback caused by the deadly root pathogen, *Phytophthora cinnamomi*. The species is now facing the threat of extinction.

Application of the fungicide Phosphite® is an effective short- to mid-term strategy for species threatened by *Phytophthora* dieback. However by 2002, establishment of new plants in a *Phytophthora*-free site was considered crucial to the long-term viability and recovery of *B. montana* and a number of other threatened species. Unfortunately, a major barrier to recovery was the lack of 'critical habitat' that was deemed disease-free within the historic range of the species. There was also insufficient stored genetic material for a comprehensive recovery effort. *Ex situ* conservation of *B. montana* was limited to a small collection of less than 300 seeds, obtained over many years and housed in DEC's Threatened Flora Seed Centre.

In 2003 a 'conservation seed orchard' was established that aimed to provide insurance against ongoing genetic erosion and to supply material for future population reconstruction, thereby taking pressure off the wild populations. The search for a suitable location away from the disease infested Stirling Range yielded a secure, disease-free site in revegetated bushland, in a lowland area on private property some 50 kilometres from the natural populations.

The establishment of the conservation seed orchard was staggered, commencing with the planting of 14 individuals of *B. montana* in 2003 (Figure 2). In 2004 a further 90 plants were added to the site and in 2005, 100 plants each of two other threatened species that co-occurred with *B. montana* in the wild were planted in the orchard. The well-drained gravelly site was deep-ripped



Figure 1. *Banksia montana* plant. Photo: Anne Cochran



Figure 2. Planting a *Banksia montana* seedling in the orchard, 2003. Photo: Anne Cochran

prior to planting, and plants were individually caged against herbivore damage. Labelling of provenance and maternal lines allows monitoring of growth and survival of individual genotypes.

Survival of *B. montana* has been high (Figure 3). By 2009, 83% of the original plantings had survived, contributing more than twice the numbers of mature plants left in the wild (37 individuals). Flowering in the orchard commenced



Figure 3. The *Banksia montana* seed orchard in 2008.
Photo: Anne Cochrane

earlier due to the less extreme environmental conditions, with almost two-thirds of individuals flowering, despite a recorded juvenile period (i.e. time span to first flowering) of 10 years in the wild. This early flowering has already allowed small quantities of seed to be collected for *ex situ* conservation, providing further insurance against species loss.

In recent years, seedlings from populations of other species currently or virtually extinct (<5 plants surviving) in the wild have been included in the seed orchard, the most notable being the Critically Endangered *Banksia brownii*. The site provides easy access for researching and monitoring biological attributes (e.g. reproductive biology), and disease hygiene can be stringently controlled. It is anticipated that research on seed orchard material may identify disease-resistant strains that can be propagated for restocking of natural populations in the future.

Conclusion

In Western Australia there are more than 400 taxa listed as Declared Rare Flora and threatened with extinction. Although protection in the wild is the highest priority for the DEC, available mitigation procedures for dieback control do not guarantee plant survival, and many critically threatened species require further intervention to prevent inevitable population decline. If the Phosphite program for *B. montana* and other susceptible species was to cease, even temporarily, these plants could be overcome by the disease. The establishment of the conservation seed orchard has proven to be a successful experiment to increase the survival prospects of *B. montana*. When all else fails, plantings like this are a feasible option.

Biotechnology and plant conservation in Australia: tissue culture and cryogenic research for *ex situ* conservation and restoration of endangered plants

Eric Bunn and Shane Turner

Botanic Gardens & Parks Authority, West Perth, WA. Email: ebunn@bgpa.wa.gov.au

What is Plant Tissue Culture?

Plant tissue culture is the process of multiplying lots of miniature plants (i.e. micropropagation), either whole plantlets or parts (e.g. shoots), under sterile conditions in closed containers (usually glass or plastic jars or tubes) on an artificial nutrient 'medium'. This nutrient medium contains minerals, organic compounds (sugar, vitamins, growth factors, plant growth compounds) and agar. The containers are autoclaved ('cooked' at high temperature and pressure) to kill all microorganisms (e.g. bacteria and fungi) that would otherwise overgrow and foul the medium if not eliminated. Starting tissue cultures similarly requires sterilising the surface of small pieces of plants (e.g. shoots or seeds) using chlorine compounds

(such as bleach or pool chlorine) and placing the sterilised plant pieces on the medium.

After a suitable time under controlled conditions of temperature and light in a culture room, the pieces of plant material begin to grow and multiply on the medium and are then cut into many pieces and placed on more tissue culture medium. In this way one shoot can be multiplied into many shoots with each successive 'sub-culture' period (usually 4 weeks), thereby producing many thousands of identical (clonal) shoots per year.

By manipulating the compounds regulating plant growth, shoots are converted to whole rooted plantlets, which can then rejoin the 'outside world'. They are transferred from the tissue culture containers into potting mixture

and grown in a glass house with fogging and temperature control until they fully adjust to the conditions outside the culture environment.

When can Tissue Culture be Used in Plant Conservation?

Plant tissue culture can be used to propagate rare or endangered plants just as it can be used to propagate horticultural plants. In the case of endangered species, tissue culture is often the only way to save them from extinction. How does this come about? There are many reasons why species become endangered, however when a species becomes extremely rare it is often very hard to obtain sufficient seeds or vegetative cutting material without endangering the remaining plants. In these cases tissue culture becomes the preferred propagation method, as only very small amounts of shoot material or even one seed can be enough to establish tissue cultures and save the species. Once established in tissue culture, a species can be maintained indefinitely, although there are some limitations to this.

Tissue culture by itself can only duplicate (clone) whatever genetic material is put into the culture system. If a species is to be saved in perpetuity the maximum amount of genetic diversity that can be saved should be saved. If insufficient genetic diversity is saved then, even if many plants are produced through tissue culture and can be returned to the wild, there may be little or no viable seed production and poor seedling recruitment from these plants. This effectively means that although the species is 'saved' (because living plants still exist), it may be unlikely to flourish even if returned to the wild in large numbers.

However not all is doom and gloom. Many species have narrow genetic diversity and are still flourishing, so this does not necessarily mean the end of the line for a particular species. There is still much to learn about this aspect and whenever we think we fully understand these issues nature continually surprises us. It simply means that great care is needed when making any assumptions regarding the fitness or otherwise of a species targeted for reintroduction. Biotechnology can now give us valuable guidance with these critical issues and the ability to react quickly, as explained in the case studies below.

Cryostorage

As culture collections increase, maintenance of them through monthly sub-culture becomes time-consuming and expensive. This prompted research into cryogenic storage technology at Kings Park, beginning in 1991. Cryostorage (cryogenic storage) is the use of liquid nitrogen to store biological samples for extended periods of time.

Cryostored tissue samples undergo no metabolic activity at the extremely low temperature of liquid nitrogen (-196°C in the liquid phase and -180°C in the vapour phase), therefore the samples remain extremely stable. Hence the

only maintenance required is the relatively small weekly replenishment of liquid nitrogen lost through vaporization from the storage containers (called dewars). Even a small to medium sized dewar can house 15-20,000 cryotubes each containing multiple cryostored samples. This is equivalent to maintaining hundreds or even thousands of culture lines at room temperature.

Two main issues with maintaining continuous culture lines at room temperature over many years are accidental contamination (usually through operator error) and somaclonal variation. The latter is the occurrence of natural variations in culture lines, such as 'sports'. Cryostorage can overcome both of these issues. The main drawback with cryogenic storage is that the skills to apply this science are only available in a limited number of laboratories in Australia. Kings Park and Botanic Garden were pioneers in applying cryogenic storage science to the conservation of endangered Australian plant species (e.g. Turner *et al.* 2001) and continue to foster cryogenic research in plant conservation.

Case Studies

The following examples illustrate the use of tissue culture and/or cryogenic storage to restore endangered plants to the wild.

Grevillea scapigera (Proteaceae)

This species is a much-cited example of the successful application of biotechnology solutions for integrated conservation of endangered plants. The species was tissue cultured (Bunn and Dixon 1992) and plants produced from the culture were used in field restoration trials. It allowed the establishment of a 'critical mass' of plants of known genetic provenance (type) to ensure outbreeding (i.e. the cross pollination of plants) and maximise viable seed production.

Inbreeding tends to occur where the plants are too closely related and eventually results in 'inbreeding depression' where each successive generation of plants is less genetically diverse than the last. Inbreeding depression had been diagnosed in *G. scapigera* by examining the genetic makeup of seedlings in the early stages of the project, and so timely remedial action was able to be taken. Cryogenic storage and *in vitro* culture of shoot tips of key plant material (of known genetic provenance) were crucial to the success of re-stocking field sites to maximise genetic variability and curtail inbreeding.

Currently *G. scapigera* is well established in field sites (see front cover) and producing copious quantities of viable seed. Importantly, seedlings are developing from the soil seed bank (Dixon and Krauss 2008).

Symonanthus bancroftii (Solanaceae)

This is an extremely rare species with only two known plants found (a single male plant located in 1996 and one female plant in 1998) since the last recorded collection in the

1940's. Despite further intensive searching, no more plants have been recorded. Both the male and female plants were established in tissue culture and plants produced (Panaia *et al.* 2000). The female plant later died in its natural habitat so tissue culturing the plants was very timely.

Micropropagated male and female plants were artificially crossed and a small amount of seed produced from which a daughter plant was obtained. This seedling in turn was successfully initiated into culture and multiplied giving three culture lines to restore plants to the wild.

Micropropagated *S. bancroftii* plants were returned to habitat sites from 2002 to 2004. About 90 plants established from 2004 plantings produced thousand of seeds from 2006 to 2008, some of which were collected. This indicated that a low genetic diversity does not always mean low seed production and/or viability. Kings Park seed scientists (D. Merritt and S. Turner) have discovered how to germinate seeds of *S. bancroftii* reliably and effectively, thus ensuring a source of seedlings when needed for field plantings.

This project is still in progress with over 200 plants and seedlings established across two field sites in remnant bushland in the Bruce Rock Shire in the Western Australia wheatbelt region. While tissue culture was crucial to this project for the first plants to be established in field sites, seed available from reintroduced plants is now the primary source of plantlets for reintroduction. It remains to be seen whether *S. bancroftii* seedlings can recruit naturally (as with *G. scapigera*) or require an environmental cue (e.g. fire or smoke) or whether there is sufficient genetic diversity for sustainable populations to persist into the future.

Emerging Techniques for Tissue Culture of Australian Plants

Somatic embryogenesis describes the artificial process of developing seed-like embryos from normal plant tissues (under special tissue cultures conditions) without the need for pollen fertilizing ovules as happens in flowers during normal seed development. The process can be scaled up in tissue culture to generate huge quantities of clonal plants for some species. The technology is relatively straightforward to apply, but takes considerable research time to optimise protocols for each species.

The Botanic Gardens and Parks Authority (BGPA) has been involved in two major studies with Australian rush and sedge species (Restionaceae and Cyperaceae) since 2003. It has achieved success with somatic embryogenesis for plant production of species in these two major groups (e.g. see Panaia *et al.* 2004).

Conclusion

Biotechnology research (plant tissue culture, storage of plant tissue in liquid nitrogen and the development of embryos from normal plant cells) greatly increases our ability to save threatened plant species from extinction and



Four-year old plants of the critically endangered Bancroft's Symonanthus (*Symonanthus bancroftii*) at their re-introduction field site, 2007. Photo: E. Bunn

enhance their survival in the wild. As more experience is gathered, existing techniques improved and new discoveries made, biotechnology remains a key instrument in the plant conservation toolbox.

Acknowledgements

Kings Park and Botanic Garden acknowledges the assistance of students, researchers, volunteers, state government agencies (DEC, Water Authority), Bruce Rock and Corrigin shires, local community conservation groups (Corrigin LandCare), and individuals and World Wildlife Fund for Nature Conservation with case studies described in this article. We also acknowledge support from RIRDC, ARC, ALCOA and Worsley Mining for projects in 2003-6 and 2006-8 on somatic embryogenesis.

References

- Bunn, E. and Dixon, K.W. (1992). *In vitro* propagation of the rare and endangered *Grevillea scapigera* (Proteaceae). *HortScience* 27: 261-2.
- Dixon, B. and Krauss, S. (2008). Translocation of the Corrigin grevillea in south Western Australia. In: Pritpal S. Soorae (ed.), *Global Re-Introduction Perspectives*. IUCN/SSC Re-introduction Specialist Group, pp 229-34.
- Panaia, M., Senaratna, T., Bunn, E., Dixon, K. and Sivasithamparam, K. (2000). Micropropagation of the critically endangered Western Australian species *Symonanthus bancroftii* (F. Muell) L. Haegi (Solanaaceae). *Plant Cell, Tissue and Organ Culture* 63(1): 23-9.
- Panaia, M., Senaratna, T., Dixon, K.W., Sivasithamparam, K. (2004). The role of cytokinins and thidiazuron in the stimulation of somatic embryogenesis in key members of the Restionaceae. *Australian Journal of Botany* 52:1-9.
- Turner, S.R., Senaratna, T., Bunn, E., Tan, B., Dixon, K.W. and Touchell, D.H. (2001). Cryopreservation of shoot tips from six endangered Australian species using a modified vitrification protocol. *Annals of Botany* 87: 371-8.

The road to recovery: the contribution of seed conservation and reintroduction to species recovery in Western Australia

Andrew Crawford and Leonie Monks

Department of Environment and Conservation, Kensington, WA. Email: andrew.crawford@dec.wa.gov.au

Introduction

Western Australia is blessed with a rich flora comprising about half the known plant taxa in Australia. Associated with this high diversity is a high level of habitat loss. These two factors combined have resulted in the south-west of Western Australia being listed as one of the world's 34 biodiversity hotspots. Currently the state has 391 plant taxa listed as being threatened (Declared Rare Flora, DRF) with a further 13 presumed to be extinct. In addition, 2429 taxa are poorly known or data deficient (Priority taxa) (Atkins 2008).

Ex situ Seed Collections

The Department of Environment and Conservation (DEC), Western Australia is the government agency responsible for the management of the above species. The Department is also responsible for developing species and area-based recovery plans to guide the recovery process. The primary goal of these recovery plans is to preserve species in their natural habitat. One of the primary conservation activities for many of these threatened species is the collection and *ex situ* storage of seeds for future use in recovery actions such as reintroduction. The Department's Threatened Flora Seed Centre (TFSC), established in 1992, is the facility responsible for collecting and storing DRF species, with the exception of orchid collections which are made by, and stored at, the Botanic Gardens and Parks Authority (BGPA).

The *ex situ* seed collections are often described as 'insurance policies' against extinction in the wild. The ultimate success of this policy lies in the use of the seeds in the augmentation, reintroduction or establishment of new populations into the wild resulting in a decreased threat level, or ideally removing the species from threat. In order to be able to claim on this insurance policy the following criteria need to be met:

- seeds must be genetically representative of the species,
- seed quantities must be adequate, and
- the seeds must be viable and germinable.

The first step in the seed conservation process is to ensure that some seeds are held in storage of all species considered at threat. Currently the TFSC's seed conservation program has collected 70% of Western Australia's Declared Rare Flora. The orchid collections held by the BGPA account for a further 10%.

Seed Collecting Strategy

The TFSC collecting strategy aims to collect seed from at least 50 individuals if a population comprises 50 or more plants, or from all individuals if the population has fewer than 50 individual plants. We aim to collect from all known populations of taxa listed as DRF, and from one representative population for Priority taxa. This latter approach has been adopted due to the sheer number of Priority taxa.

Ideally seeds from each individual within a collection are kept separate. This adds value to the collection, as it allows maternal lines to be balanced in a reintroduction or opens up the possibility of genetic work being carried out. Many DRF have been labelled in the field. By keeping individuals separate, we can track how many seeds have been obtained from each. This allows us to specifically target individuals which are not represented, to increase the diversity of the collections.

How Many Seeds to Collect?

Theoretically collections should be sufficiently large to allow for the assessment of initial seed quality, monitoring of viability over time and for utilisation when required. TFSC uses a target of 10,000-20,000 seeds for its collections. However due to the rare nature of most species TFSC deals with, this target is often not achieved. For instance, of the 332 collections of Critically Endangered taxa held by the TFSC in 2007, half had less than 1000 seeds and only five exceeded 10,000 seeds (Cochrane *et al.* 2007).

Where seed numbers are limited, achieving meaningful measures of initial seed viability and then monitoring this viability over time becomes problematic. At the TFSC, 10% of a collection is deemed to be a reasonable proportion that can be utilised to achieve these outcomes. The remainder is stored until required for use in species recovery.

The target of 10,000-20,000 seeds may seem high, however if the overall aim is to reintroduce self-sustaining populations that are viable in the long term, then many times this number of seeds may be required (see Cochrane *et al.* 2007 for examples). Much can still be achieved even with limited numbers of seeds. For example, *Grevillea batrachioides* is known from only one population of 63 plants and had less than 600 seeds in storage. A translocation has resulted in 97 currently surviving plants, most of which have flowered and set seed.



Grevillea batrachioides. Photo: Andrew Crawford

Testing Seed Quality

Seed viability is an important measure of seed quality, however germination is arguably more useful when utilisation of the collection is considered. Seed dormancy may pose a limitation on the number of plants produced from a given seed lot until the mechanism of dormancy breaking is better understood. Viability tests can be subjective (e.g. tetrazolium test) and often destructive (e.g. cut test). In contrast, germination tests can produce seedlings that may be used for either reintroductions or research (e.g. susceptibility to *Phytophthora cinnamomi*, Shearer *et al.* 2004).

Ongoing monitoring of the viability/germinability of seed collections is important to ensure that viable material is available when finally required. Monitoring the viability of collections becomes problematic when collection sizes are small (see Guerrant and Fiedler (2004) for a discussion of this issue). The TFSC has adopted a sequential acceptance sampling approach (Whitehead and Marek 1985) to better utilise available seeds. Re-testing is part of a risk management strategy. Where recent germination results are available, some confidence can be taken in the viability of a given seed accession. If a recent germination result is not available, then confidence in the viability of a collection can come into doubt. In the absence of enough seeds to re-test, the TFSC's management assumption is that viability will have decreased and further collections will need to be made.

Re-testing of collections at the TFSC was initially carried out after one year in storage, then at five years, 10 years and every 10 years thereafter (when adequate seeds were available). Crawford *et al.* (2007) showed that a high proportion of collections maintained viability in the short (<5 years) or medium (5-12 years) term. In cases where viability decline was seen, it often appeared linked to collection quality rather than the species being inherently short lived. This result reinforces the importance of making high quality seed collections for *ex situ* storage. Initial re-tests are now carried out after five years of storage, or if seeds are limiting, after ten years.

Using Seed for Reintroductions

The true measure of the success of a seed conservation program is the impact on the status of the species in the wild through the utilisation of seeds in reintroduction programs. The DEC currently has 34 seed-based reintroductions underway which represent 9% of the DRF taxa. A further 13 reintroductions have been initiated using other propagation methods, which in combination with the seed-based reintroductions represent 12% of the DRF.

Seeds collected by the TFSC are the primary source of material utilised in the reintroduction program. During the reintroduction process losses inevitably occur through the germination, nursery propagation and planting out phases. For example, for every 100 seeds of *Grevillea humifusa*, 76 adult plants were produced at the reintroduction site. In contrast, just seven adult plants were produced on average from 100 seeds of *Daviesia bursarioides* (Cochrane *et al.* 2007). This feedback loop provides valuable information about how many seeds are needed to produce one mature individual, thereby assisting in prioritising species for further seed collection.



Seedlings of *Grevillea humifusa* have successfully established at this translocation site. Photo: Andrew Crawford

Table 1. Status of seed-based reintroductions implemented by the Department of Environment and Conservation in Western Australia, 2008.

| Status of reintroduction | Number of taxa |
|--|----------------|
| Number of seed based reintroductions | 34 |
| Number taxa that have established | 29 |
| Number of taxa that have failed to establish | 5 |
| Number of taxa that have established and reproduced | 23 |
| Number of taxa that have established but not reproduced | 3 |
| Number of taxa that have established but reproduction is unknown | 3 |
| Number of taxa that have established, reproduced and recruited a second generation | 4 |
| Number of taxa that have established, reproduced but not recruited a second generation | 19 |
| Number of taxa that have established, reproduced and a recruited third generation | 0 |
| Number of taxa that have established, reproduced and not recruited a third generation | 23 |

Reintroduction Success

Reintroductions aim to establish or maintain viable self-sustaining populations in the wild. Ultimately, we hope that reintroduced populations will assist in ensuring that the taxa are secure enough to be removed from the threatened list. A series of criteria are set, as part of the reintroduction planning process, to assess success. Over the past decade these criteria have been refined. They vary slightly between taxa with different life forms, but essentially assess whether the reintroduced plants establish (measured by survival and growth), reproduce (measured by production of flowers, fruit and viable seeds) and recruit a second and subsequent generations (measured by counting naturally recruited seedlings). Based on these criteria 29 of the 34 seed-based reintroductions have resulted in the successful establishment of plants at new or current locations (Table 1). Of these, 23 taxa have successfully reproduced, but just four taxa have naturally recruited a second generation.

Five reintroductions have failed to establish. Four of these were direct-seeded with little or no germination recorded, and the fifth failed due to dry conditions immediately after planting. When dealing with limited numbers of seeds the use of seedlings appears to be the best option for establishing new populations.

The taxa that established but have not reproduced were all planted in the last 18 months. These species are long-lived perennials so reproductive maturity is not yet expected. It is not surprising that many taxa have not recruited second or subsequent generations when the long generation times are taken into account. In addition, many of these taxa recruit only after disturbance and disturbance events have not occurred during the life time of the reintroductions.

Conclusion

Ex situ seed conservation underpins Western Australia's threatened flora reintroduction program which has successfully established new populations of 7% of the state's Declared Rare Flora. These new populations have greatly increased the number of plants in the wild,

however, with most it is still too early to tell whether the new populations will persist long-term without further intervention. There have been some failures, but much has been learnt from these 'experiments' in reintroduction that can be applied in future reintroductions, hopefully to improve their success. The important point is that seeds in *ex situ* storage do not necessarily guarantee the survival of a species *in situ*.

Many challenges are faced in recouping the insurance policy represented by seeds in *ex situ* storage. This is a challenge that is being embraced in Western Australia by DEC. The department hopes that these efforts will be rewarded in the near future, with these actions starting to positively alter the conservation status of the state's most threatened plant species.

References

- Atkins, K.A. (2008). *Declared Rare and Priority Flora List*. Department of Conservation and Land Management, Western Australia.
- Cochrane, J.A., Crawford, A.D. and Monks, L.T. (2007). The significance of *ex-situ* seed conservation to reintroduction of threatened plants. *Australian Journal of Botany* 55: 356-61.
- Crawford, A.D., Steadman, K.J., Plummer, J.A., Cochrane, J.A. and Probert, R.J. (2007). Analysis of seed bank data confirms suitability of international seed storage standards for the Australian flora. *Australian Journal of Botany* 55: 18-29.
- Guerrant, E.O. and Fiedler, P.L. (2004). Accounting for sample decline during *ex-situ* storage and reintroduction. In: E.O. Guerrant, K. Havens and M. Maunder (eds), *Ex-situ plant conservation. Supporting species survival in the wild*. Island Press, Washington DC USA, pp 365-86.
- Shearer, B.L., Crane, C.E. and Cochrane, J.A. (2004). Quantification of the susceptibility of the native flora of the South-west Botanical Province, Western Australia, to *Phytophthora cinnamomi*. *Australian Journal of Botany* 52: 435-43.
- Whitehead, J. and Marek, P. (1985). A FORTRAN program for the design and analysis of sequential clinical trials. *Computers and Biomedical Research* 18: 176-83.

Black Gum, a nationally threatened tree of upland New South Wales and Victoria

Steve Douglas

Ecological Surveys & Planning, Albury, NSW.

Description and Conservation Status

Eucalyptus aggregata is a small- to medium-sized tree endemic to the Central and Southern Tablelands of New South Wales and the Central Highlands of Victoria (where it occurs as a disjunct population) (Hill 2002). It is commonly known as Black Gum because of its relatively distinctive, often dark-coloured bark.

Black Gum occurs in grassy woodlands on alluvial soils in moist sites along creeks on broad, cold and poorly-drained flats and hollows (DSE 2004; Field 2007; Hill 2002). In New South Wales, it commonly occurs with Candlebark (*Eucalyptus rubida*), Ribbon Gum (*E. viminalis*) and Snow Gum (*E. pauciflora*), with a grassy understorey of River Tussock (*Poa labillardieri*) (Field 2007). The population in Victoria occurs primarily in 'frost pockets' in which Black Gum may dominate or co-dominate with Swamp Gum (*Eucalyptus ovata*) (DSE 2004).

Black Gum is listed as Endangered under the *Flora and Fauna Guarantee Act 1988* (Vic.) and has been nominated as Vulnerable under the *Threatened Species Conservation Act 1995* (NSW). At the time of writing, it was not listed under the *Environment Protection and Biodiversity Conservation Act 1999* (C'th) but warrants at least a Vulnerable status. It is also part of an ecological community nominated as Endangered in New South Wales (see Crooks *et al.* 2008).

Threats

Habitat loss and fragmentation

Black Gum occurs naturally in relatively arable, grassy and moist landscapes and thus is likely to have been subject to considerable habitat loss and degradation. Large areas of apparently suitable habitat, which occur between remnant populations, no longer support this species and show evidence of tree removal, thinning, and the replacement of the native vegetation with exotic grasses or pine forests. The main land uses in its habitat are pastoralism and plantation forestry (*Pinus radiata*), though rural-residential and industrial uses also occur. Growth in demand for 'lifestyle' properties within areas around a three hours drive from Sydney, Canberra and Melbourne, along with decreasing viability of livestock grazing, is likely to fuel increased rates of subdivision of rural holdings. This may pose a threat to Black Gum through further clearing and fragmentation of remnants. As many remnants are now restricted to road verges, road maintenance (especially road widening) is a notable threat.

Small population size

Field (2007) surveyed all of the officially recorded populations of Black Gum in New South Wales and found a further 15-20 populations, making 110 in total. Of all populations, about 90% occur as scattered trees on heavily grazed, privately-owned land that showed little or no recruitment of Black Gum seedlings. Several populations additional to those listed by Field (2007) have been recorded, but all were very small (Douglas, personal observation). The population of Black Gum in Victoria is reported to consist of about 9000 plants (see DSE 2004). However, the largest Black Gum populations in New South Wales only contain around 700-1000 adults, which is very small compared to the largest populations of other *Eucalyptus* species (10,000s-100,000s) (Field 2007). The small size of the populations is magnified by them often being separated by large areas of cleared, degraded and grazed habitat, and by their naturally fragmented or at least patchy occurrence as a result of their narrow habitat requirements.

Genetic 'swamping'

Field (2007) examined the threat posed by hybridisation between Black Gum and Candlebark, Ribbon Gum and Mountain Gum (*E. dalrympleana*). He concluded that hybridisation was "quite a conservation concern for this species, as in some of the small remnant stands, as many as 35% of the seed crops are hybrids. Furthermore, most populations have many adult hybrids which are fertile with as high as 25% of adults also being of mixed ancestry and there is evidence of substantial backcrossing and introgression" (Field, pers. comm.). Hybridisation between Black Gum and Swamp Gum (*E. ovata*) has been observed in the Victorian population, although such hybrids are rare (see DSE 2004).

Weed invasion/competition

In the Victorian population, DSE (2004) note that "A wide range of woody and herbaceous weeds compete with Black Gum seedlings for space, light and water. The overall effect is usually to restrict rather than prevent regeneration. Woody weeds include Blackberry (*Rubus discolor*), Gorse (*Ulex europaeus*), willows (*Salix species*), Hawthorn (*Crataegus monogyna*) and English Broom (*Cytisus scoparius*)".

Much the same can be said for the populations in New South Wales. The moderately fertile and at least seasonally moist habitat of Black Gum is particularly prone to weed invasion/establishment, especially by introduced pasture plants such as *Phalaris* spp. and Cocksfoot (*Dactylis*

glomerata) (Field 2007; Douglas, personal observation). With most Black Gum populations being small, linear remnants within road reserves (Field 2007), the extent and intensity of weed invasion in these often un- or poorly-managed lands is a significant threat.

Poorly reserved and not readily reserved

Small populations of Black Gum occur in Tallaganda, Morton, Yanununbeyan and Blue Mountains national parks, and in Turallo Nature Reserve. Such occurrences are usually on the periphery of these reserves, or in the case of Yanununbeyan, are restricted to a former inholding that was apparently subject to a grazing lease that predates the reserve's gazettal.

Any remedy to Black Gum's poor representation in the conservation estate is largely reliant on appropriate covenanting or purchasing of privately-owned land by government or non-government conservation bodies. Two of the larger remnants exist within publicly-owned travelling stock reserves, but such reserves and their associated stock routes are at risk of being sold on the basis that they are no longer needed for pastoral purposes. Whilst there are numerous remnants within publicly-owned road reserves, these are unlikely to be able to be added to effective conservation areas, and they often suffer from severe edge-effects due to them frequently being the only remnant vegetation in what are otherwise largely cleared pastoral landscapes.

Climate change

Black Gum is apparently a remnant from cooler climates in geological time when it is presumed that the already fragmented pre-European occurrences were connected (DSE 2004). Natural and anthropogenic climatic change may threaten this species, particularly in relation to more frequent, lengthy and severe droughts; more frequent and severe fires; higher intensity rainfall events that may worsen erosion of its habitat; and more frequent and intense severe winds which could fell or significantly damage trees. The latter is a particular concern in areas where there is no recruitment and where general habitat stresses are high. Climatic warming may pose a particular threat to this species. Field (2007) attributed the deaths of a significant proportion of saplings in recent years to the on-going drought in south-eastern Australia.

Conclusion

Black Gum is threatened by a range of anthropogenic factors, but appears to have been in long-term decline due to non-anthropogenic climate change (i.e. gradual warming since the last ice age). A naturally fragmented distribution, small and increasingly fragmented populations, poor reservation, difficulty in achieving adequate reservation, weed invasion, hybridisation, further land clearing and climate change are all of concern. The species warrants recognition, protection and recovery under state and federal legislation.



Black Gum (Eucalyptus aggregata) vegetation in Blue Mountains National Park. Photo: Steve Douglas

References

- Crooks, J., Rehwinkel, R., Treweek, A. and Baines, G. (2008). The conservation value and reservation status of the Snow Gum, Black Sallee, Candlebark and Ribbon Gum Grassy Woodlands of south-eastern NSW. *Australasian Plant Conservation* 17(1): 24–25.
- DSE (2004). *Action Statement No. 84. Black Gum Eucalyptus aggregata*. Department of Sustainability & Environment (Victoria), Melbourne.
- Field, D.L. (2007). *The importance of ecological factors in determining the pattern of interspecific hybridization in fragmented landscapes of Eucalyptus aggregata*. Unpublished PhD Thesis, University of Wollongong.
- Hill, K.D. (2002). Eucalyptus. In G.J. Harden (ed.), *Flora of New South Wales*, Revised Edition, Volume 2. University of NSW Press, Sydney.

A new population of Minyon Quandong (*Elaeocarpus sedentarius*) from northern New South Wales

Lui Weber

Sinclair Knight Merz, South Brisbane, Qld. Email: Lxweber@skm.com.au

Species Information

Minyon Quandong (*Elaeocarpus sedentarius*) is a threatened mid-sized tree known only from simple notophyll vine forest / warm temperate rainforest in northern New South Wales. It is endemic to the Mt Warning volcanic caldera and was thought to be extinct until 1992, when it was rediscovered on the Nightcap Range north of Lismore. The species is listed as Endangered under the *Environment Protection and Biodiversity Conservation Act 1999* (C'th) and Vulnerable under the *Threatened Species Conservation Act 1995* (NSW). Presently it is known from five populations totalling approximately 570 adults and seedlings (NSW National Parks & Wildlife Service 2003). Four of the populations are on the Nightcap Range with the fifth being on the nearby Koonyum Range near Mullumbimby (Rossetto *et al.* 2008).

This article describes the discovery of a new outlying population on the other side of the Mt Warning caldera and discusses the implications for the conservation of the species.

A recent study of Minyon Quandong found significant genetic and morphological differences between the four Nightcap Range populations and the single Koonyum Range population (Maynard *et al.* 2008). The study also noted no evidence of recent gene flow between populations on the two ranges despite a distance of only five kilometres separating them.

An indication of the extreme relictual nature of the species is grounded in the fact that highland populations of the closely related *E. blephoceras* from Papua New Guinea are less distinct in morphometric analysis (i.e. based on morphological characters) from Minyon Quandong than they are from lowland *E. blephoceras* (Maynard *et al.* 2008). Genetic analysis of Minyon Quandong has revealed that potentially this species and its close relative from New Guinea may not be correctly placed in the genus *Elaeocarpus* and may represent an ancient lineage that is basal to *Elaeocarpus*, *Aceratium* and related genera (Crayn *et al.* 2006). Further research is needed to clarify this finding.

New Population

While recording detailed vegetation plots during a survey for the Department of Environment and Climate Change, New South Wales in 2007, I located a large individual of Minyon Quandong in the Tomewin area on the McPherson Range north of Murwillumbah. This location lies about 30 kilometres to the north of the nearest known population on the Koonyum Range. The specimen cannot possibly have been planted, as the plant has four stems 15-30 cm diameter at breast height and its age is estimated to be 50-100 years. As the species was thought to be extinct until rediscovered in 1992, it was not propagated until after that date.

All previously known populations are found on acid volcanic rhyolite derived soils supporting warm temperate rainforest dominated by Coachwood (*Ceratopetalum apetalum*), or occur in nearby Brushbox (*Lophostemon confertus*) forest with a rainforest understorey (Rossetto *et al.* 2008).

The new site is in a gully rainforest on soils derived from metasediment with Brush Box and secondary species such as Pencil Cedar (*Polyscias murrayi*) and White Bollygum (*Neolitsea dealbata*). The site is situated at 100 metre elevation in a south-east facing gully, and as such is considerably outside the known habitat parameters for the species, having a different soil parent rock and



Flowers on the tree at the new location for Minyon Quandong. Photo: Lui Weber



Leaves and the distinctive fibrous fruits of the endangered Minyon Quandong. Photo: Lui Weber

occurring at a lower altitude. More typical habitats for Minyon Quandong with rhyolite soil, higher elevations and Coachwood occur further west along the range where the new population was located. This area warrants further survey to locate additional individuals of the species.

Casual observations suggest that the new individual of Minyon Quandong differs morphologically in having broader leaves and fewer lateral veins than the Nightcap and Koonyum Range populations. Samples have been submitted for genetic and morphometric analysis by the authors of the previous study.

It is likely that this individual, due to its physical and ecological isolation from other populations, will prove to offer unique genetic traits not found in the two other populations, and so increase the species' chances of survival in the long term.

References

- Crayn, D.M., Rossetto, M. and Maynard, D.J. (2006). Molecular phylogeny and dating reveals an Oligo-Miocene radiation of dry-adapted shrubs (Tremandraceae) from rainforest tree progenitors (Elaeocarpaceae) in Australia. *American Journal of Botany* 93: 1168–82.
- Maynard, D., Crayn, D., Rossetto, M., Kooyman, R. and Coode, M. (2008). *Elaeocarpus sedentarius* sp. nov. (Elaeocarpaceae)—morphometric analysis of a new, rare species from eastern Australia. *Australian Systematic Botany* 2: 192–200.
- NSW National Parks & Wildlife Service (2003). *Recovery Plan for the Elaeocarpus sp. Rocky Creek (syn E. sp. 2 'Minyon')*. NSW National Parks & Wildlife Service, Hurstville.
- Rossetto, M., Kooyman, R., Sherwin, W. and Jones, R. (2008). Dispersal limitations, rather than bottlenecks or habitat specificity, can restrict the distribution of rare and endemic rainforest trees. *American Journal of Botany* 95: 321–9.

Is fencing enough to conserve Western Australia's wheatbelt woodlands?

Rachel Standish¹, Suzanne Prober², Chris Curnow³ and Jeff Richardson⁴

¹School of Environmental Science, Murdoch University, WA.

²CSIRO Sustainable Ecosystems, Wembley, WA. Email: Suzanne.Prober@csiro.au

³WWF-Australia, Wembley, WA.

⁴Department of Environment and Conservation, Bentley, WA.

Less than a century ago, the slopes and valley floors of the gently undulating landscape of hinterland south-western Australia were clothed with eucalypt woodlands of extraordinary floral diversity. By the 1960s, about 10 million hectares of woodlands had been cleared for agriculture and the 'wheatbelt' came into existence.

Today, as we battle the environmental, social and economic costs of such rapid and widespread clearing, the scattered remnant woodlands are precious for the biological diversity they retain, and the lessons they can teach us about the processes that enable plants and animals to function on such a dry and infertile landscape. Maintaining these processes is crucial to ensuring the flora and fauna can survive into the future as the effects of global change become more apparent.

Since the early 1990s there has been a determined effort to fence remnant wheatbelt woodlands from grazing livestock. Early on, people assumed that fencing would be enough to ensure the conservation of relatively intact woodlands and the recovery of degraded woodlands. Despite the considerable investment in fencing programs, these assumptions have not been tested. To address this gap, and supported by funding from the Avon Catchment Council, we have instigated a study to evaluate the benefits of fencing herb-rich York Gum–Jam (*Eucalyptus loxophleba*–*Acacia acuminata*) woodlands. The team decided to focus on this woodland type as it represents one of the more heavily cleared and least conserved eucalypt woodlands.

The study uses an 'over-the-fence' comparison, whereby fenced and unfenced sections of the same remnant are surveyed for their native floral diversity and cover, weed cover, soil condition and tree recruitment (Figure 1). We will then relate this information to that gleaned from the land owners about the age of the fence and historical factors (e.g. disturbance by fire) that might account for



Figure 1. Over-the-fence comparisons are being used to measure outcomes of fencing programs. Tree recruitment, especially of Jam wattle, is evident in the area on the right, which was fenced in 1998. Photo: Suzanne Prober

recovery or lack thereof. In addition, floral composition, tree recruitment and soil condition of fenced remnants will be compared with a selection of relatively undisturbed 'reference sites', to indicate the extent of recovery towards the natural state of the woodlands.

So far the team, with the assistance of Georg Wiehl and Steve Zabar (CSIRO), have visited 60 farms and surveyed over 40 remnants. Georg is currently compiling the data and checking the identity of the plants collected in the field. The ultimate aim is to characterise sites where fencing is enough to prevent further decline of the woodland and, conversely, identify those sites where further restoration will be necessary.

Acknowledgement

We wish to thank the numerous land owners for their generous hospitality and for sharing their stories with us, and the Avon Catchment Council for funding support.

Endangered plant discovered by seven year old boy

Doug Beckers

Department of Environment and Climate Change, Sydney. Email: doug.beckers@environment.nsw.gov.au

Seven year old Zac Bissett has astounded National Parks and Wildlife Service (NPWS) officers with his recent discovery of the threatened mint bush *Prostanthera askania* in Bouddi National Park. Found only on the Central Coast of New South Wales, *P. askania* is listed as endangered under the *Threatened Species Conservation Act 1995* (NSW) and has not previously been found in any conservation reserve.

Local Macmasters Beach residents Zac, and his great aunt Maureen Findlay, discovered the plant while walking in bushland in Bouddi National Park. Zac noticed a nice 'mint' smell so he and his great aunt investigated. As the mint bush was not one that Maureen had seen before she referred to a plant identification book.

Discovering that the find was likely to be something of significance, Maureen approached NPWS Bush Regeneration Co-ordinator Deb Holloman to confirm the identification. NPWS Acting Regional Manager Jenni Farrell said that staff were amazed upon hearing of Zac's discovery.

"For a child so young to show this interest and enthusiasm for the natural environment is just fantastic. With the species listed as endangered this find is highly significant and is actually the first record of an endangered plant in Bouddi National Park."

Over 500 individual plants have been recorded at the location, making it an important population and extending the known range of the species. The species previously has been recorded within the catchments of Ourimbah Creek, Narara Creek, Dog Trap Gully, Chittaway Creek and Berkeley Creek and Erina and Fires Creeks, with a geographic range of 12 kilometres.

Botanists from the NSW Herbarium met with Zac and Maureen while visiting the site a few weeks later, but the surf was up and Zac couldn't stay long.



Prostanthera askania. Photo: Doug Beckers



Zac Bissett looking at his discovery. Photo: Doug Beckers

Research Roundup

Byrne, M., Yeates, D.K., Joseph, L., Kearney, M., Bowler, J., Williams, M.A.J., Cooper, S., Donnellan, S.C., Keogh, J.S., Leys, R., Melville, J., Murphy, D.J., Porch, N. and Wyrwoll, K.-H. (2008). **Birth of a biome: insights into the assembly and maintenance of the Australian arid zone biota.** *Molecular Ecology* 17(20): 4398-417.

Gibbons, P., Briggs, S.V., Ayers, D.A., Doyle, S., Seddon, J., McElhinny, C., Jones, N., Sims, R. and Doody, J.S. (2008). **Rapidly quantifying reference conditions in modified landscapes.** *Biological Conservation* 141(10): 2483-93.

Hayward, M.W. and Kerley, G.I.H. (2008). **Fencing for conservation: Restriction of evolutionary potential or a riposte to threatening processes?** *Biological Conservation* 142(1): 1-13.

Hovenden, M.J., Williams, A.L., Kongstad Pederson, J., Vander Schoor, J.K. and Wills, K.E. (2008). **Elevated CO₂ and warming impacts on flowering phenology in a southern Australian grassland are related to flowering**

time but not growth form, origin or longevity. *Australian Journal of Botany* 56(8): 630-43.

Jarrad, F.C., Wahren, C.-H., Williams, R.J. and Burgman, M.A. (2008). **Impacts of experimental warming and fire on phenology of subalpine open-heath species.** *Australian Journal of Botany* 56(8): 617-29.

Joseph, L.N., Maloney, R.F., O'Connor, S.M., Cromarty, P., Jansen, P., Stephens, T. and Possingham, H.P. (2008). **Improving methods for allocating resources among threatened species: the case for a new national approach in New Zealand.** *Pacific Conservation Biology* 14: 154-8.

Kolb, A. (2008). **Habitat fragmentation reduces plant fitness by disturbing pollination and modifying response to herbivory.** *Biological Conservation* 141(10): 2540-9.

Stevens, T.C., Puotinen, M.L. and Whelan, R.J. (2008). **Powerline easements: ecological impacts and contribution to habitat fragmentation from linear features.** *Pacific Conservation Biology* 14: 159-68.

Information Resources and Useful Websites

Environmental Weeds of Australia

Sheldon Navie and Steve Adkins

University of Queensland

Price: AUD\$59.00 (excluding GST and postage)

This is an interactive DVD that includes a Lucid3 key to just over 1,000 major environmental weeds of Australia, detailed descriptions of weed species, links to website information, a detailed cross linked glossary and thousands of images. The publishers describe it as "an invaluable resource to all those involved with research, training and management of environmental weeds in Australia, especially State and local weed control officers, Bushcare and Landcare volunteers". A review of the DVD can be found in the Australian Systematic Botany Society (ASBS) Newsletter No. 137, December 2008, pages 18-21 (available at <<http://www.anbg.gov.au/asbs/newsletter/newsletter-pdf/08-dec-137.pdf>>). The DVD can be ordered through the University of Queensland online shop at <<http://shop.cbbit.uq.edu.au/ProductDetails.aspx?productID=189>>.

Invasive Species Council

<http://www.invasives.org.au/home.html>

The Invasive Species Council is an NGO set up in 2003 to create awareness about and lobby against all types of invasive species. It recently published the first issue of

its electronic bulletin *Double Trouble*, which includes articles on the potential for an explosion of weeds in Victoria following the recent extensive bushfires there, the potential impact of the root rot fungus *Phytophthora* on Western Australia's biodiversity hotspots, invasive species and climate change, and many more topics. Those interested can subscribe to the e-bulletin at the web address for Issue 1, <<http://invasives.org.au/doubletrouble/doubletrouble1.html>>.

Lippia Management Manual

National Lippia Working Group

The National Lippia Working Group has recently published this manual, which collates historical work on Lippia (*Phyla canescens*) and current and ongoing activities of landholders and the research community. Lippia is an invasive weed originally introduced as a garden ornamental plant. It has now infested about 5.3 million hectares of the Murray-Darling Basin where it threatens native vegetation, soils and biodiversity, especially in floodplain and riparian areas. The manual contains sections on the distribution, impacts and threat of Lippia in Australia (Section 1), its biology, ecology and genetics (Section 2), management to control the weed (Section 3) and 24 case studies (Section 4). The document can be downloaded from <<http://www.namoi.cma.nsw.gov.au/37.html>>.

Information Resources and Useful Websites (cont.)

Bitou Bush Management Manual

M.A. Winkler, H. Cherry and P.O. Downey (eds)

Department of Environment and Climate Change, 2008

A weed of national significance, Bitou Bush (*Chrysanthemoides monilifera* subsp. *rotundata*) affects coastal ecosystems in south-east Queensland, New South Wales and north-east Victoria. The manual describes the biology of and threat posed by Bitou Bush (Section 1), planning and pre-control considerations (Section 2), guidelines on how to manage the species in different habitats (Section 3), control methods (Section 4), linking control with restoration (Section 5) and how to monitor control progress (Section 6). Section 7 contains 11 case studies, while Section 8 provides additional information including safety and other legal requirements, useful contacts and education and awareness materials. The manual can be downloaded from <<http://www.weeds.org.au/WoNS/bitoubush/>>.

Botanical Resource Centre

Australian National Botanic Gardens, Canberra

Open daily, 9.30 am – 4.30 pm

The Australian National Botanic Gardens (ANBG) has opened a botanical resource centre containing a reference herbarium of nearly 5000 plants from the ACT and surrounding south-east region (from Sydney to the Victorian border and almost as far inland as Wagga). It also contains a small library of botanical reference books. Visitors can use the centre to help identify many native plants or weed species in the region, using interactive computer keys, hard-copy keys, microscopes and the public reference herbarium. Trained facilitators are available on Wednesdays and Sundays 1 pm – 4 pm to help visitors learn to use the facility. For further information, contact the ANBG Visitor Centre on (02) 6250 9540.

Mistletoes

<http://www.anbg.gov.au/mistletoe/>

This website from the Australian National Botanic Gardens explores the world of mistletoes and provides a wealth of information about them. Topics include defining a mistletoe, dispersal and germination, mistletoes as food plants for butterflies, whether mistletoes show cryptic mimicry of their hosts, mistletoes growing on other mistletoes, the evolutionary origin of mistletoes and their biogeography in the southern hemisphere, mistletoes and bushfires, and mistletoes in folk legend and medicine. It contains a checklist of mistletoe species currently recognised in Australia together with the host species each has been recorded on, plus, for many species, photos of their flowers and/or fruit. If you're not a mistletoe enthusiast to start with, you will be after spending time in this site!

A field manual for surveying and mapping nationally significant weeds, 2nd Edition

I. McNaught, R. Thackway, L. Brown and M. Parsons

Bureau of Rural Sciences, Canberra, July 2008

This manual explains standardised, systematic procedures for collecting core weed infestation data for mapping of weeds of national significance. It can be downloaded from <<http://affashop.gov.au/product.asp?prodid=13948>>.

Dictionary of the fungi, 10th Edition

P.M. Kirk, P.F. Cannon, D.W. Minter and J.A. Stalpers

CSIRO Publishing / CABI

Price: \$175.00

Published in November 2008, this new edition contains more than 21,000 entries and provides the most complete listing available of generic names of fungi, their families and orders, their attributes and descriptive terms. For each genus, the authority, the date of publication, status, systematic position, number of accepted species, distribution, and key references are given. Diagnoses of families and details of orders and higher categories are included for all groups of fungi. Further information is available at <<http://www.publish.csiro.au/nid/20/pid/5998.htm>>.

Other recently published books

A field guide to plants of the Barkly Region Northern Territory

By Jenny Purdie, Chris Materne and Andrew Bubb, Barkly Landcare and Conservation Association, 2008. Price \$35.00. Further information: www.barklylandcare.org.au/5-Publications.html

Floodplain woodland plants of north east Victoria: identification of native weeds and practical weed management for bush regeneration projects

By Helen Curtis and Peter Curtis, Wangaratta Urban Landcare Group, 2008. Price \$16.00. Further information: <http://northeast.landcarevic.net.au/wangaratta-urban>

Geology of the Canberra Region

Produced by the Geological Society of Australia, and includes a 1:100,000 scale geological map Geology of the Australian Capital Territory, a GIS CD (geological map plus additional data), and guidebook A Geological Guide to Canberra Region and Namadgi National Park. Further details: <http://www.gsa.org.au/publications/index.html>

Woodland to weeds: southern Queensland Brigalow Belt, 2nd edition

By Nita C. Lester, Annerly, Brisbane. Price \$65 + postage. Further information: <http://www.nitalester.com/>

ANPC Forum

Australian Network for Plant Conservation, 2nd National Forum *Minding our own biodiversity: conservation on private land*

Thursday 30 April – Friday 1 May 2009

Halls Gap, The Grampians, Victoria

Our second national forum will focus on conservation outside the formal reserve system, whether on small blocks, production properties, large landscape level efforts or cross-tenure projects. It will highlight the people and places involved and investigate the incentives and support available.

The forum will include presentations, case studies, facilitated discussion sessions and field visits to project sites. Many landholder presenters will give us the benefit of their on-ground experience.

This is your opportunity to participate, learn, contribute, debate and move this essential component of biodiversity conservation forward.

**Don't miss this timely forum
REGISTER NOW!**

Registrations close Friday 17 April

Further information, program, registration:

<http://www.anpc.asn.au/conferences.html>

or contact the ANPC office

on 02-6250 9509.

Private land is the key to linking biodiversity conservation across the landscape

Conferences and Workshops

International Plant Propagators Society – Joint Meeting of the Australian & NZ Chapters

Fruitful 42' South

14-17 May 2009

Hobart, Tasmania

Topics include propagation methods, potting mixes for propagation, irrigation systems, Millenium Seed Bank work, invasive plants and the nursery industry, and native plant breeding. One day of the program will have been set aside for tours to a range of nurseries and sites around Hobart.

Further information: <http://sites.google.com/site/ippstas/Home/2009-tasmania-conference>

Centre for Historic Research, National Museum of Australia

Barks, Birds & Billabongs: the American-Australian Scientific Expedition to Arnhem Land 1948 Remembered

6-10 July 2009

Canberra, ACT

This international symposium will investigate the expedition's significant and often controversial legacy. It will be organised around three core themes: Histories, Legacies and Continuing Traditions, and reflect the interdisciplinary makeup of the expedition itself. Particular emphasis will be placed on Indigenous perspectives. A diverse range of stimulating and innovative speakers will give presentations appealing to both specialised scholars as well as to the general public. The symposium will be launched at a public lecture delivered by a high profile speaker.

Further information: http://www.nma.gov.au/barks_birds_billabongs

Conferences and Workshops (cont.)

**The International Association for Ecology (INTECOL)
10th International Congress
*Ecology in a Changing Climate:
Two Hemispheres – One Globe***

16-21 August 2009

Brisbane, Qld

Hosted by The Ecological Society of Australia and The New Zealand Ecological Society, conference participants will explore how global climate change has impacted, and will further impact, ecosystems and their vital services to human communities. They will examine unique features of ecosystems in the southern and northern hemispheres but look for common elements in a search for solutions to this looming problem. Symposia will represent all scales of ecology from individual organisms to landscapes, and report on a diversity of ecosystems—from marine to freshwater aquatic systems, and from arid to rainforest and polar to tropical terrestrial ecosystems.

Further information: <http://www.intecol10.org>

**Society for Ecological Restoration International
(SERI)**

**19th World Conference on Ecological Restoration
*Making Change in a Changing World***

23-27 August 2009

Perth, WA

SERI world conferences aim to provide a critical platform to assist in defining the principles of restoration, understanding goals and milestones, debating what ecosystem functions to measure and closing the gap between the science of restoration ecology and the practice of ecological restoration.

Themes to be included in the 19th conference include:

- restoration ecology in a changing world
- social and cultural aspects of restoration
- restoration at the landscape scale
- ecological restoration of ecosystems
- ecological restoration of disturbed sites
- ecological restoration within urban areas
- ecological restoration of threatened species, populations and habitats
- management and technical aspects
- disturbance ecology and management.

Further information: <http://www.seri2009.com.au/pages/home.html>

**Charles Darwin University & NT Government
*Charles Darwin: Shaping our Science,
Society & Future***

22-24 September 2009

Darwin, NT

The year 2009 marks the 200th anniversary of the birth of Charles Darwin and the 150th anniversary of his work *The Origin of Species*. Through a combination of meticulous observation and innovative thinking, Darwin developed an explanation for the incredible variety of living things: that evolution is driven by natural selection. The symposium will provide an opportunity to appreciate, debate, and even challenge Darwin's findings, and will bring together an exciting range of speakers from around the globe.

Further information: <http://www.cdu.edu.au/cdss>

Advanced notice of other conferences:

Australian Plants in the 21st Century

Conference of the Association of Societies for Growing Australian Plants, 26 September – 2 October 2009, Geelong, Vic.

Further information: <http://asgap2009.apsvic.org.au/>

Plan(e)t Priority: Regional Reality

2009 Botanic Gardens of Australia and New Zealand (BGANZ) Congress, 8-10 October 2009, Mackay, Qld.

Further information: <http://www.bganz.org.au/congress.html>

Systematic botany: from science to society

Australian Systematic Botany Society (ASBS) Conference, 29 November – 6 December, 2009

University of New England, Armidale. The conference will highlight discovery, analysis and synthesis in plant diversity research and its impact on society, and will include a workshop on national accreditation of providers of biological identification.

Further information: Jeremy Bruhl (jbruhl@une.edu.au)

Threatened species and ecological communities newly listed under the EPBC Act

In January 2009 the federal Minister for the Environment, Heritage and the Arts announced that the following eight plant species had been listed as Critically Endangered: *Hibbertia* sp. Bankstown (R.T. Miller & C.P. Gibson s.n. 18/10/06), *Thelymitra* sp. Kangaloon (D.L. Jones 18108) (Kangaloon Sun Orchid), *Thelymitra cyanapicata* (Dark-tipped Sun Orchid), *Caladenia intuta* (Ghost Spider-orchid), *Cassinia tegulata*, *Notelaea ipsviciensis* (Cooneana Olive), *Phebalium distans* (Mt Berryman Phebalium) and *Reedia spathacea* (Reedia).

The Minister also listed the following two ecological communities as Critically Endangered:

- Gippsland Red Gum (*Eucalyptus tereticornis* subsp. *mediana*) Grassy Woodland and Associated Native Grassland, and
- Natural grasslands on basalt and fine-textured alluvial plains of northern New South Wales and southern Queensland.

The following ecological communities were listed as Endangered:

- Alpine Sphagnum Bogs and Associated Fens,
- Natural Grasslands of the Queensland Central Highlands and the northern Fitzroy Basin, and
- Weeping Myall Woodlands.

Further details about these species and ecological communities can be found through www.environment.gov.au/biodiversity/threatened/index.html.



Endangered Weeping Myall (*Acacia pendula*) woodland.
Photo: Rosemary Purdie



Critically Endangered Natural grasslands on basalt and fine-textured alluvial plains of northern New South Wales and southern Queensland. Photo: Rosemary Purdie

ANPC Corporate Members

ANPC acknowledges the support of the following corporate members

Albury Botanic Gardens, NSW

Greening Australia, VIC

Australian National Botanic Gardens, ACT

Redland Shire Council, QLD

Botanic Gardens of Adelaide, SA

Royal Botanic Gardens Melbourne, VIC

Botanic Gardens Trust, NSW

Royal Tasmanian Botanic Gardens, TAS

Department of Environment and Conservation, WA

Sydney Olympic Park Authority, NSW

Dept of Sustainability and Environment, VIC

University of Melbourne, Burnley Campus, VIC

ANPC Major Sponsors

GOLD SPONSORS



Australian National
Botanic Gardens, ACT



Centre
for Plant
Biodiversity
Research



NSW
Environmental
TRUST

OTHER SPONSORS

Seeking APC sponsors

If you and your organisation value receiving *Australasian Plant Conservation* (APC), please consider sponsoring a future issue. Any contribution helps defray the publication costs, and such donations are tax deductible. You'll be able to include a short note about yourself or your organisation, and what you do, in the issue you sponsor. Phone the ANPC Office (02 6250 9509) or email the APC Editor, Rosemary Purdie (Rosemary.Purdie@environment.gov.au) if you'd like to sponsor an issue or get more information.

Plant Germplasm Conservation in Australia

Strategies and Guidelines for developing, managing and utilising *ex situ* collections



FULLY REVISED EDITION

Edited by: C.A. Offord and P.F. Meagher

ANPC in partnership with MSB Australian Partners



Chapter 1: Introduction, including Plant Conservation in the international and Australian contexts

Chapter 2: Germplasm conservation options and major considerations

Chapter 3: Seed and Vegetative material collection

Chapter 4: Seed Banking

Chapter 5: Seed Germination and Dormancy

Chapter 6: Tissue Culture

Chapter 7: Cryopreservation

Chapter 8: Living Plant Collections

Full of practical case studies to demonstrate principles of germplasm conservation in Australia

A handy companion to the 'Guidelines for the Translocation of Threatened Plants in Australia' (Second edition)

SUPPORTED BY: ANBG, LWA, BGT (NSW), DEH (SA), RGB KEW

Available from September 2009

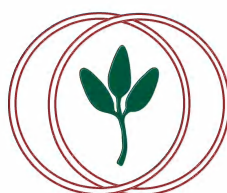
Refer to Website for Order form

<http://www.anpc.asn.au/books.html> or from the ANPC Office (details below)



Australian Government
Australian National Botanic Gardens

exchange
national vegetation knowledge service



Australasian Plant Conservation

BULLETIN OF THE AUSTRALIAN NETWORK FOR PLANT CONSERVATION INC

For further information contact:

Australian Network for Plant Conservation
GPO Box 1777
Canberra ACT 2601, Australia

Ph: + 61 2 6250 9509
Fax: + 61 2 6250 9528
Email: anpc@anpc.asn.au
Website: <http://www.anpc.asn.au>